University of Waterloo

Waterloo, Ontario, Canada
N2L 3G1

Faculty of Science
Department of Biology
519/885-1211

(11)

November 22, 1982

Dear Colleague,

Don't forget the 11th Plant Development Workshop to be held in the Biology Building at the University of Waterloo, Saturday November 27th.

A timetable is enclosed.

See you on the 27th.

Carol Peterson

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PROGRAM

11th PLANT DEVELOPMENT WORKSHOP

UNIVERSITY OF WATERLOO

9:00 - 10:00 AM	REGISTRATION
10:00 - 10:15	Covariate measurements increase precision of lettuce growth and development studies. D.P. Ormrod.
10:15 - 10:30	Root growth under field conditions. Marian Stypa and M.H. Miller.
10:30 - 10:45	A natural auxotroph of a predatory hyphomycete. K. Zachariah and J.R. Victor.
10:45 - 11:00	A structural study of the interaction between the ectomycorrhizal fungus Pisolithus tinctorius (Pers.) Coker and Couch and Pinus strobus L. roots. Y. Piche, R.L. Peterson, M.J. Howarth and J.A. Fortin.
11:00 - 11:30	COFFEE BREAK
11:30 - 11:45	A correlated study of the structural and permeability changes in onion root cortical walls in response to wounding. Greg Moon and Carol A. Peterson.
11:45 - 12:00 N	Siliceous fibres occurring in the lemma and palea of grasses. A. G. Sangster, M.J. Hodson and D. Wynn Parry.
12:00 - 12:15	Leaf surface wax of corn. T.W. Blaker.
12:15 - 12:30	Gibberellins in sugar maple: methodology and phenological patterns. E.M. Zeng, R.L. Legge and E.B. Dumbroff.
12:30 - 2:30	LUNCH AND DISCUSSION OF POSTER.
poster	Morphogenesis in Betula papyrifera. I. Short shoot organogenesis. A.D. Macdonald, D.H. Mothersill and J.C. Caesar.
2:30 - 2:45	Cellular differentiation in Lotus corniculatus callus. M.J. Howarth, R.L. Peterson and D.T. Tomes.
2:45 - 3:00	Kinetin and caffeine: antagonists or mimics? D. Davidson and E. Pertens.
3:00 - 3:15	Chromosomal elimination in caffeine-induced bimitotic cells of a diploid interspecific hybrid of Hordeum: the influence of cycloheximide. George Wheatley.

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3:15 - 3:30	Developmental process in both vegetative and floral shoots of the hybrid grape cultivar Ventura. U. Posluszny and R.E. Subden.
3:30 - 3:45	Developmental changes in embryos of sugar maple during the stratificaiton - germination process. C.Y. Shih, Carol A. Peterson and E.B. Dumbroff.
3:45	WORKSHOP FOR PARTICIPANTS: METHODOLOGY FOR TESTING FERMENTED GRAPE EXTRACT.

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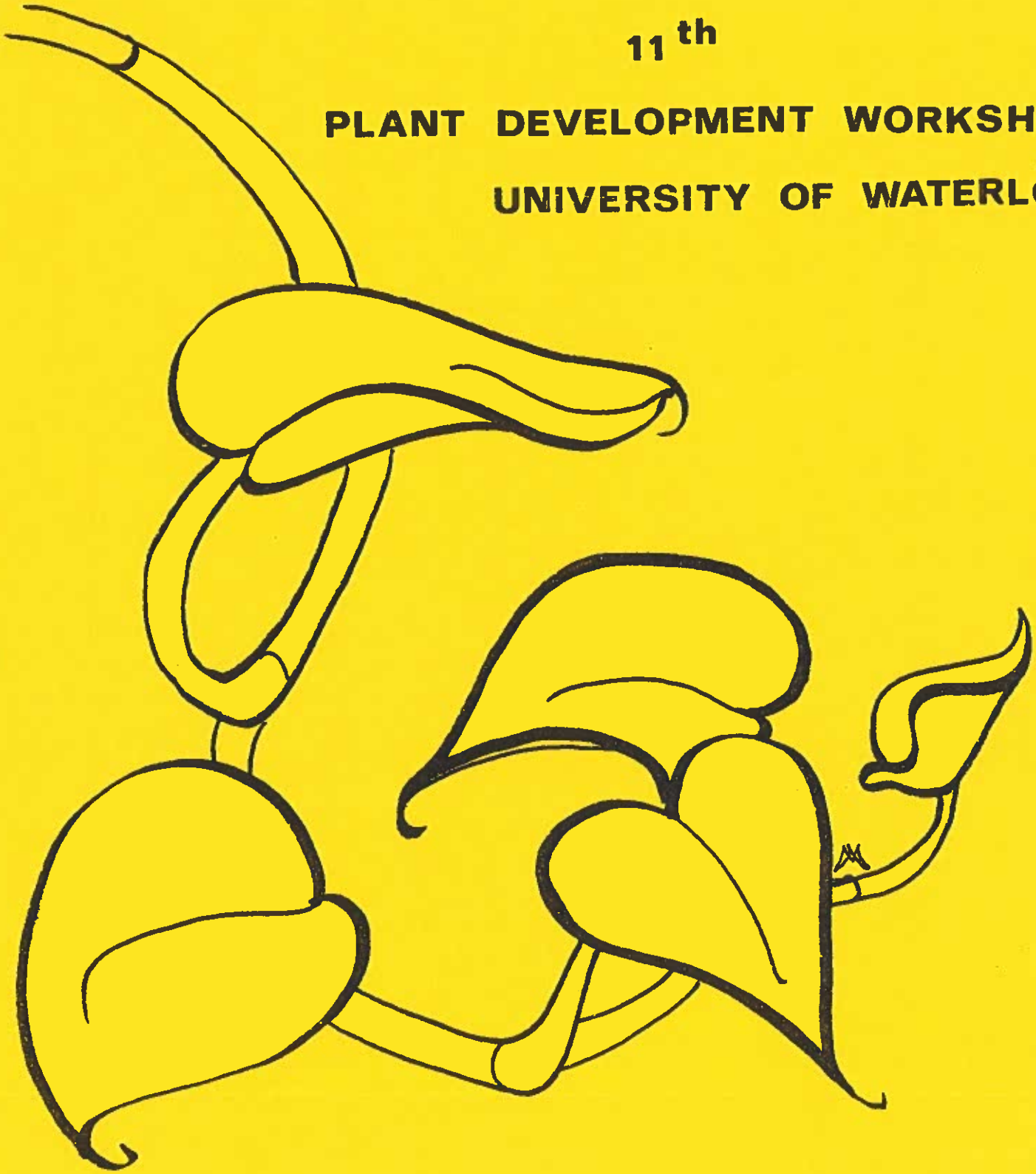
11th PLANT DEVELOPMENT WORKSHOP

UNIVERSITY OF WATERLOO



PROGRAM and ABSTRACTS

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9:00-10:00 A.M. REGISTRATION

10:00-10:15 COVARIATE MEASUREMENTS INCREASE PRECISION OF LETTUCE GROWTH AND DEVELOPMENT STUDIES

D.P. Ormrod, University of Guelph.

Lettuce plants (Lactuca sativa L. cv Grand Rapids) were exposed to clean air or ozone and/or sulphur dioxide for 6 hours at 14 days from seeding in controlled environments using base-line environmental conditions. High plant-to-plant variability in harvest data resulted in statistical analyses that identified few significant treatment effects on growth and development. The utilization of non-destructive covariate measurements prior to pollutant exposures followed by analysis of covariance of harvest data markedly increased the ability of the experiment to detect treatment effects. Planar area estimates were useful as covariates for leaf area, fresh weight and dry weight at harvest 3 or 7 days after exposure. Plastochron index (PI), a measure of plant development, was useful as a covariate for PI at harvest.

10:15-10:30 CORN (ZEA MAYS L.) ROOT GROWTH UNDER FIELD CONDITIONS

M.L. Stypa. U. of Waterloo, M.H. Miller. U. of Guelph.

A series of treatments was imposed to remove surface and subsurface soil strength, fertility, and moisture as limitations. Two levels of fertility were used under irrigated and non-irrigated conditions. In one of the treatments, the soil was removed to a depth of one meter and replaced with an artificial growth medium consisting of soil-peat-perlite (3:1:1 by volume). Root length was measured three times during the 1981 growing season. The above ground dry matter production was monitored throughout the growing season. Soil bulk density and soil strength were also measured.

Although the rooting depth in the artificial medium was greater than that in the soil treatments, the total root growth was less. Thus the lower bulk density and higher porosity of the artifical medium did not stimulate root proliferation. Irrigation had no effect on dry matter production, or final yield. Above ground total dry matter and grain yield were increased by fertility rates above those recommended by soil test. In this study, there was no evidence that physical properties of the soil at the Elora Research Station constituted a significant limitation to root growth or yield of corn during the 1981 growing season.

10:30-10:45 Title: A natural auxotroph of a predatory hyphomycete

K. Zachariah & J. R. Victor. Biology Department, University of Waterloo, Waterloo, Ontario N2L 3G1

Predatory fungi efficiently trap and consume their nematode prey; yet surprisingly they have quite ordinary nutritional needs. They grow well on simple defined media, even on distilled water agar. We describe a strain of Arthrobotrys dactyloides from forest soil which has an absolute requirement for two amino acids and a partial need for six others. Such natural auxotrophs are rare in the fungi and it is possible they exist in the wild as heterokaryons with prototrophs. Attempts to synthesize heterokaryons of this kind yielded two prototrophic sectors which when subcultured produced small yellow apothecia in clusters on a compact stroma. Sections reveal a dense hymenium of very fine paraphyses, but no asci have been detected at any stage of development yet. So, it is probable that the selective advantage of the auxotroph lies in its ability to initiate the sexual cycle. Other features of its physiology such as ability to sporulate and predatory performance will be described.

10:45-11:00 A structural study of the interaction between the ectomycorrhizal fungus Pisolithus tinctorius (Pers.) Coker & Couch and Pinus strobus L. roots

Y. Piche[1], R.L. Peterson[2], M.J. Howarth[2], and J.A. Fortin[1].
[1]Department D'Ecologie et Pedologie, Universite Laval, Ste.-Foy, Quebec; [2]Department of Botany and Genetics, University of Guelph, Guelph, Ontario.

Stages in ectomycorrhizal development between the fungus Pisolithus tinctorius, and short roots of Pinus strobus were followed in growth pouches. Short roots from pre-inoculation through Hartig net formation were processed for light and electron microscopy. Fungal hyphae approaching the surface of roots have Thiery-positive substances in their modified walls and in lomasomes, indicating the possibility of extracellular polysaccharide secretion. Hyphae grow between and beneath the flattened, tannin-filled superficial root cells and subsequently into the intercellular region of the cortex. A Hartig net several hyphae wide is formed, isolating the cortical cells from each other. Plasmodesmata were not observed in these cortical cells. Middle lamella material is always present around the intercellular hyphae, suggesting limited digestion by the hyphae. Cortical cell cytoplasm becomes necrotic in regions of mature Hartig net formation.

11:00-11:30 COFFEE BREAK

11:30-11:45 A CORRELATED STUDY OF THE STRUCTURAL AND PERMEABILITY CHANGES IN ONION ROOT CORTICAL WALLS IN RESPONSE TO WOUNDING

Greg Moon and Carol A. Peterson. Biology Department, University of Waterloo, Waterloo, Ontario.

Onion roots were wounded by scoring them with a needle 8 to 12 cm behind the root tips. The plants were maintained in nutrient solution. Various histochemical, dye penetration, and transmission electron microscope (TEM) investigations were carried out immediately following wounding and for six days thereafter. Starting two days after wounding, pit callose was apparent. At the same time, the cortical cells and intercellular air spaces adjacent to the wound stained positively for lipids using sudan IV, resisted concentrated sulphuric acid digestion and were autofluorescent under ultraviolet light. Taken together, these tests indicate that suberin was deposited. The quantity of suberin appeared to increase until day four. Penetration studies using Calcofluor White M2R, an apoplastic dye, showed that the wound was completely sealed off to apoplastic transport two days after wounding. Using TEM, no suberin lamellae were observed in the cortical cell walls surrounding the wound. The intercellular air spaces were filled with an electron-dense material. Thus, in response to wounding, nonlamellar suberin is deposited in the cortical cell walls and air spaces surrounding the wound and is confluent with the suberin normally present in the hypodermis.

11:45-12:00 N Siliceous fibres occurring in the lemma and palea of grasses. A.G. SANGSTER, M.J. HODSON and D. WYNN PARRY. Division of Natural Science, York University, Toronto, and University College of North Wales, U.K. Inflorescence bracts are well known sites of heavy silicon accumulation in grasses (Poaceae). In various Phalaris species, the heaviest silicon accumulation detected was associated with the outer (abaxial) epidermis of the lemma. Preferential deposition sites were located in the cell walls of trichomes (macrohairs), tbe walls and lumina of long cells, and external to the epidermis, forming a thin layer of extracellular sheet silica. Occasional silicification of internal tissues occurred either as elongated, dendriform fibres, resulting from the infilling of lumina and pits of hypoderm fibres, or as rows of spongy mesophyll cells. Dimensions and silicon levels recorded for the various siliceous structures indicated that the macrohairs and the friable sheets formed by silicification of the abaxial epidermal cells were a potential source of sharp, elongated, siliceous fibres. These fibres may have a possible carcinogenic significance.

12:00-12:15 LEAF SURFACE WAX OF CORN

Timothy W. Blaker, Department of Plant Sciences,
University of Western Ontario
LONDON, Ontario

A gas chromatographic study of the leaf surface wax composition of corn leaves has shown that there are differences between the leaves present on a seedling and the leaves present on a mature plant. These differences can be seen directly, in the traces from the gas chromatograph, and can be characterized numerically as well. Statistical analyses have been used to describe the nature of the change from one composition to the other.

12:15-12:30 GIBBERELLINS IN SUGAR MAPLE: METHODOLOGY AND PHENOLOGICAL PATTERNS

E.M. Zeng, R.L. Legge and E.B. Dumbroff
Department of Biology, University of Waterloo, Waterloo, Ontario, Canada, N2L 3G1

Although functional gibberellins are apparently critical for the transition of woody perennials from the dormant to the active state, they have received comparatively little attention since they are, in many respects, the most difficult group of plant hormones to analyze. In the present work, the gibberellin content in young stems of sugar maple was examined during an entire vegetative cycle in an attempt to define the correlation between endogenous gibberellin activity and several physiological events that characterize dormancy, quiescence and growth. A procedure for the extraction, separation and measurement of endogenous free (ethyl acetate soluble) and conjugated (n-butanol soluble) gibberellins in sugar maple was developed. Both free and conjugated gibberellins were detected in February during the terminal phase of the chilling requirement when the buds were entering the quiescent state. A second peak of activity in the acidic, n-butanol-soluble gibberellins extended from bud-break during the second week of May through the rapid flush of growth lasting well into June. Analysis of selected samples showed that high levels of conjugated abscisic acid may have masked the activity of conjugated-gibberellins.

12:30-2:30 LUNCH AND DISCUSSION OF POSTER

poster MORPHOGENESIS IN BETULA PAPYRIFERA. I. SHORT SHOOT ORGANOGENESIS.
Macdonald, A.D., D.H. Mothersill and J.C. Caesar. Dept. of Biology, Lakehead University, Thunder Bay, Ont. P7B 5E1

White birch possesses long and short shoots. Potential short shoot apices arise in the axils of the more proximal foliage leaf primordia of long shoot buds in mid-July, year n. In year n + 1 one rudimentary leaf, 2 - 3 foliage leaves and 3 rudimentary leaf primordia are formed from early May to mid-July. Internode extension is minimal. Flushing occurs in spring of year n + 2. The stipules of the last-formed rudimentary leaves develop as bud scales, 2 - 3 foliage leaves and 3 rudimentary leaf primordia form from early May to mid-July to complete a short shoot terminal bud. Flushing occurs in spring of year n + 3 and the sequence may repeat. A short shoot apex may become transformed into a female inflorescence apex in July of year n + 1, or of any subsequent year (designated year f). At transformation 2 transitional leaves are formed after the foliage leaf primordia, the apex rapidly expands and bracts arise low on the apical dome. Flowering occurs with leaf emergence in spring, year f + 1. In July of year f, in a potential reproductive short shoot bud, a vegetative apex arises in the axil of one or both foliage leaf primordia, and 2 - 3 rudimentary leaf primordia are initiated rapidly. 2 - 3 primordial foliage leaves and 3 rudimentary leaf primordia arise in year f + 1 to complete a short shoot axillary bud on a flowering short shoot. This sequence may repeat.

2:30-2:45 Cellular differentiation in Lotus corniculatus callus.

M.J. Howarth, R.L. Peterson and D.T. Tomes. Department of Botany and Genetics, University of Guelph (MJH & RLP), and Pioneer Hi-Bred International, Johnston, Iowa, USA (DTT).

Lotus callus cultures were studied in an attempt to determine at the cellular and subcellular levels, what morphological changes precede and accompany differentiation. Small clumps of homogenized callus were plated onto a medium containing benzyladenine which was known to induce differentiation in this system. Initially, callus was yellowish and consisted of large, vacuolated cells with deposits of starch. Marked changes occur in these cells; peripheral and endogenous meristematic areas are initiated, giving rise to shoots, and either groups of tracheary elements or roots, respectively. Roots developed within five days while shoot apical meristems with leaf primordia formed by day nine. Many of the cells surrounding meristematic areas developed suberin lamellae in their walls, while others, both within and on the periphery of meristematic areas, accumulated phenolic substances. Morphological observations tend to support the view that physiological isolation of cultured tissue may precede differentiation.

2:45-3:00 KINETIN AND CAFFEINE: ANTAGONISTS OR MIMICS?

D. Davidson and E. Pertens. Biology Dept., McMaster University, Hamilton, Ontario.

Kinetin (6-furfurylaminopurine) and caffeine (1,3,7-trimethylxanthine) are both purine derivatives. The inhibitory effects of caffeine on proliferating cells have been discussed at previous meetings of this group. These effects are exhibited, in root meristems, by inhibition of: 1) RNA and protein synthesis, 2) cell and nuclear growth, 3) rate of progress through the cell cycle and 4) cytokinesis, which resulting in the formation of binucleate cells. Responses 1-3 are stimulated, in normal systems, by kinetin. We have determined, therefore, the ability of kinetin to reverse the effects of caffeine on pea root meristems. The response of meristems treated only with kinetin, however, has revealed close parallels with the response to caffeine. A 3 hr treatment with 10 ppm kinetin, for example, produced 1.27% binucleate cells. By 9 hr this frequency had risen to 1.43% (each determination is based on 3000 cells: 1000 cells scored from each of 3 roots). Furthermore, at 9 hr, 24% of the binucleate cells were in mitosis. Thus, kinetin and caffeine share one inhibitory property: they prevent cytokinesis. Kinetin, however, stimulates the binucleate cells to complete a mitotic cycle in 9 hr, or less. The consequences of these effects of kinetin will be discussed in relation to the stability of proliferating cell populations.

3:00-3:15 Chromosome elimination in caffeine-induced bimitotic cells of a diploid interspecific hybrid of Hordeum: the influence of cycloheximide

George Wheatley, University of Guelph, Crop Science Dept.

The experiment presented in this paper was undertaken to examine whether cycloheximide (CH), an inhibitor of protein synthesis which has been previously shown to increase chromosome elimination in mitotic cells in root meristems of a diploid interspecific hybrid between H. vulgare and H. bulbosum, acts on processes inherent to the hybrid nucleus or on processes within the cytoplasm to induce elimination.

A 4h treatment with 0.1% caffeine was used to induce a subpopulation of binucleate cells in root meristems of the diploid interspecific hybrid. Later, when the binucleate cells were in bimitosis, the roots were treated for 1h with 10 ug/ml CH and then allowed to recover for 1 and 2h after the treatment. Over the course of the experiment observations were made on the frequency of occurrence of bimitotic cells exhibiting chromosome elimination in neither, one or both mitoses and on the frequency of mitotic cells in the mononucleate cell subpopulation in these roots which exhibited chromosome elimination.

Examination of the bimitotic cells at the end of the 1h CH treatment indicated that the two mitoses of a bimitotic cell appeared to undergo elimination independently of each other: a much higher percentage of bimitotic cells exhibited elimination in only one mitosis compared to the percentage of bimitotic cells in which both mitoses exhibited elimination. During the recovery period relatively high percentages of bimitotic cells in which both mitoses exhibited elimination were observed suggesting that both mitoses may have been undergoing elimination in concert. Comparisons of the occurrence of elimination in the bimitotic cells, on a per mitosis basis, with the response of mitoses in the mononucleate subpopulation in the same roots suggest however that the mitoses of the treated hybrid roots underwent a similar response to the inhibitor regardless of whether they share a common cytoplasm in bimitotic cells or whether they occur in separate cells. From this, it would appear that CH affects the two mitoses of a binucleate cell differentially and that CH most likely acts on processes inherent to the hybrid nucleus to increase chromosome elimination.

3:15-3:30 Developmental process in both vegetative and floral shoots of the hybrid grape cultivar Ventura

U. Posluszny and R.E. Subden. Department of Botany and Genetics, University of Guelph.

Although grapes are amongst the oldest cultivated crops, very little critical scientific work has been done on the development of the vegetative and floral shoots. As in other plants with a vine-like habit the branching in the grape shoot is highly unorthodox. The axillary buds (usually 2-6 in each axil) are formed early in the summer of the year preceding bud break. The bud possesses partially developed embryonic structures of next year's flowers, tendrils and shoots. All three of these structures can develop from a structure called an anlage. What determines the kind of structure (flower, tendril or shoot) the anlage will form and the sequence on the stem is unknown. Within the overwintering bud numerous hair-like outgrowths develop, surrounding the meristematic tissue. The development and possible function of these hair-like outgrowths is currently being investigated.

3:30-3:45 DEVELOPMENTAL CHANGES IN EMBRYOS OF SUGAR MAPLE DURING THE STRATIFICATION-GERMINATION PROCESS

C.Y. Shih, C.A. Peterson and E.B. Dumbroff
Biology Department, University of Waterloo, Waterloo, Ontario, N2L 3G1

Changes in the anatomy, morphology and stored food reserves of stratifing (5°C) sugar maple embryos were followed from a strongly dormant state through germination. The correlation of cell division and cell elongation with radicle protrusion was also investigated. An effective clearing technique was devised which allowed detailed examination of cellular components and an in situ view of the vascular system in the whole embryo. First germination was observed on day 38, but the first phloem and xylem elements were noted on the 20th and 40th days respectively. A rapid uptake of water induced a large increase in the fresh weight of the emerging axes; however, cell division was not detected until 5 days after germination when axes were more than 10 mm long. Although no changes in the amount of protein, starch or lipid were noted during stratification, a more scattered arrrangement of oil droplets in the cells was observed, and an apparent disappearance of birefringence of starch grains and protein bodies in the axes characterized the beginning of food mobilization at the onset of germination.

3:45 WORKSHOP FOR PARTICIPANTS: METHODOLOGY FOR TESTING FERMENTED GRAPE EXTRACT.